## Records of limno-terrestrial tardigrades (Tardigrada) from Sarnena Gora

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**Abstract**. There is no previously published data on tardigrades found in Sarnena Gora region. During the study period a total of 13 samples were collected. Specimens from genera *Echiniscus* C.A.S. Schultze, 1840, *Milnesium* Doyère, 1840, *Isohypsibius* Thulin, 1928, *Ramazzottius* Binda & Pilato, 1986, *Adropion* Pilato, 1987 and family Macrobiotidae Thulin, 1928 were discovered.

Key words: Sarnena Gora, tardigrades, distribution.

#### Introduction

Tardigrades, also known as water bears, are micrometazoans  $(500 - 1200 \ \mu\text{m})$  that occupy a diversity of niches in freshwater, marine, and terrestrial habitats. As a phylum, tardigrades have a worldwide distribution. Some species with broad ecological requirements are still considered to be cosmopolitan, whereas others with more narrow tolerances are rare or endemic (Nelson *et al.* 2015).

Data on Bulgarian tardigrades is limited and for now it is not an object of international research interest. Recent studies have been conducted by Kaczmarek *et al.* (2011), Georgiev (2016), Geogiev & Kenderov (2017) and Morek *et al.* (2019) with the latter describing new species for science. Older records reported by Iharos (1961, 1973, 1982) should be confirmed by modern methods of tardigrade research.

There is no previously published data on tardigrades found in Sarnena Gora region. During the study period a total of 13 samples were collected. Specimens from the genera *Echiniscus* C.A.S. Schultze, 1840, *Milnesium* Doyère, 1840, *Isohypsibius* Thulin, 1928, *Ramazzottius* Binda & Pilato, 1986, *Adropion* Pilato, 1987 and family Macrobiotidae Thulin, 1928 were discovered.

The presented data in the paper is a starting point for further research of the diversity of Bulgarian tardigrades, including Sarnena Gora region. Long term research and more samples are needed for better quantitative and ecological data on water bears in the country.

#### TARDIGRADA

#### **Material and Methods**

Samples from ground, tree and rock substrate were collected and later soaked in water for 12 to 24 hours. After that period, the water containing tardigrades was examined under stereomicroscope and light microscope. Species were identified to genus level using published description and modern keys. The taxonomy follows Degma *et al.* (2019).

#### **Results and Discussion**

Tardigrades were established in 61.5% of the samples. A total of 165 tardigrades were extracted with *Milnesium* species found in 47% of all samples. It's worth to be noted that from a single sample of algae in a spring *Milnesium* specimen with claw configuration [2-3][2-2] was extracted and *Adropion* sp. exuvia was found.

#### Class Heterotardigrada Marcus, 1927

#### Family Echiniscidae Thulin, 1928 Genus Echiniscus C.A.S. Schultze, 1840 Echiniscus merokensis Richters, 1904

Material collected on 16.05.2015, North of Stara Zagora City, near Tabashka River; Habitat: bush and grass vegetation; Sample: moss *Grimmia* sp. on limestone rocks; GPS: N42 27 20.5 E25 37 58.4, 287 m a.s.l.; Number of specimens discovered: 3 (without B and D appendages); Material collected on 30.05.2015, Northwest of Zmeevo vill.; Habitat: bush and grass vegetation; Sample: moss *Grimmia* sp. on limestone rocks; GPS: N42 30 34.8 E25 35 46.8, 472 m a.s.l; Number of specimens discovered: 2.

#### Echiniscus testudo (Doyère, 1840)

Material collected on 16.05.2015, North of Stara Zagora City, near Tabashka River; Habitat: bush and grass vegetation; Sample: moss *Grimmia* sp. on limestone rocks; GPS: N42 27 20.5 E25 37 58.4, 287 m a.s.l.; Number of specimens discovered: 8; Material collected on 30.05.2015, Northwest of Zmeevo vill.; Habitat: bush and grass vegetation; Sample: moss *Grimmia* sp. on limestone rocks; GPS: N42 30 34.8 E25 35 46.8, 472 m a.s.l; Number of specimens discovered: 11.

Remark: Ten unidentified specimens from genus *Echiniscus* were also found from the mentioned localities.

#### Class Apotardigrada Guil, Jørgensen & Kristensen, 2019

#### Family Milnesiidae Ramazzotti, 1962 Genus *Milnesium* Doyère, 1840 *Milnesium* sp.

Material collected on 27.02.2015, Samarsko Zname National Monument, Stara Zagora; Habitat: Park; Sample: moss from alley; GPS: 42 433 922N 25 653 094E, 226 m a.s.l; Number of specimens discovered: 6 (Sex:  $\Im$ ; Claws configuration: [2-3]-[3-2] and [2-3]-[3-3]). Material collected on 16.05.2015, North of Stara Zagora City, near Tabashka River; Habitat: bush and grass vegetation; Sample: moss *Grimmia* sp. on limestone rocks; GPS: N42 27 20.5 E25 37 58.4, 287 m a.s.l.; Number of specimens discovered: 6 (Sex:  $\Im$ ; Claws configuration: [2-3]-[3-2]). Material collected on 30.05.2015, Northwest of Zmeevo vill.; Habitat: bush and grass vegetation; Sample: moss *Grimmia* sp. on limestone rocks; GPS: N42 30 34.8 E25 35 46.8, 472 m a.s.l; Number of specimens discovered: 16 (Sex:  $\Im$ ; Claws configuration: [2-3]-[3-2], [2-3]-[3-3] and [3-3]-[3-3]). Material collected on 28.03.2018, Ostra Mogila; Habitat: Spring; Sample: algae; GPS: 42 27 10.8N 25 28 27.5E, 368 m a.s.l.; Number of specimens discovered: 2 (Sex:  $\Im$ ; Claws configuration: [2-3]-[2-2]).

### TARDIGRADA

#### Milnesium cf. berladnicorum Ciobanu, Zawierucha, Moglan & Kaczmarek, 2014

Material collected on 01.03.2015, Krairechen park, Stara Zagora; Habitat: Park; Sample: *Bierkandera* sp. from *Populus* sp.; GPS: N42 26 31.5 E25 38 19.5, 228 m a.s.l.; Number of specimens discovered: 1 (Sex:  $\Im$ ; Claws configuration: [2-3]-[2-2]).

# Milnesium inceptum Morek, Suzuki, Schill, Georgiev, Yankova, Marley & Michalczyk, 2019

Published by Morek *et al.* (2019): Material collected on 25.08.2015, Shanovo vill., Kazanlak Valley, North Slope of Sarnena gora; Habitat: Village; Sample: *Grimmia* sp. moss from brick wall; GPS: 42 33 27N 25 37 51E, 300 m a.s.l.; Number of specimens discovered: 1 (Sex:  $\Im$ ; Claws configuration: [3-3]-[3-3]).

#### Class Eutardigrada Richters, 1926

# Family Isohypsibiidae Sands, McInnes, Marley, Goodall-Copestake, Convey & Linse, 2018

#### Genus Isohypsibius Thulin, 1928

#### Isohypsibius cf. prosostomus Thulin, 1928

Material collected on 30.05.2015, Zmeevo Quarry; Habitat: Oak forest; Sample: moss from *Quercus* sp.; GPS: N42 30 09.2 E25 36 33.8, 427 m a.s.l.; Number of specimens discovered: 3.

#### Family Macrobiotidae Thulin, 1928

#### Genus Macrobiotus C.A.S. Schultze, 1834

#### Macrobiotus sp.

Material collected on 27.02.2015, Samarsko Zname National Monument, Stara Zagora; Habitat: Park; Sample: moss from alley; GPS: 42 433 922N 25 653 094E, 226 m a.s.l; Number of specimens discovered: 2. Material collected on 16.05.2015, North of Stara Zagora City, near Tabashka River; Habitat: bush and grass vegetation; Sample: moss *Grimmia* sp. on limestone rocks; GPS: N42 27 20.5 E25 37 58.4, 287 m a.s.l.; Number of specimens discovered: 3.

#### Genus Mesobiotus Vecchi, Cesari, Bertolani, Jönsson, Rebecchi & Guidetti, 2016

Material collected on 16.05.2015, North of Stara Zagora City, near Tabashka River; Habitat: bush and grass vegetation; Sample: moss *Grimmia* sp. on limestone rocks; GPS: N42 27 20.5 E25 37 58.4, 287 m a.s.l.; Number of specimens discovered: 1.

Remark: Eggs are required for detailed taxonomic identification of specimens from genus *Macrobiotus* and *Mesobiotus*, which were not found in the collected samples.

#### Family Hypsibiidae Pilato, 1969 Genus Adropion Pilato, 1987

## Adropion scoticum (Murray, 1905) species complex

Material collected on 28.03.2018, Ostra Mogila; Habitat: Spring; Sample: algae; GPS: 42 27 10.8N 25 28 27.5E, 368 m a.s.l.; Number of specimens discovered: 2 (Sex: ♀; Claws configuration: [2-3]-[2-2]).

#### Genus Ramazzottius Binda & Pilato, 1986 Ramazzottius cf. oberhaeuseri (Doyère, 1840)

Material collected on 16.05.2015, North of Stara Zagora City, near Tabashka River; Habitat: bush and grass vegetation; Sample: moss *Grimmia* sp. on limestone rocks; GPS: N42 27 20.5 E25 37 58.4, 287 m a.s.l.; Number of specimens discovered: 1; Material collected on 30.05.2015, Northwest of Zmeevo vill.; Habitat: bush and grass vegetation; Sample: moss *Grimmia* sp. on limestone rocks; GPS: N42 30 34.8 E25 35 46.8, 472 m a.s.l. Number of specimens discovered: 16.

Sarnena Gora is one of the many regions in Bulgaria that lack records of tardigrade fauna. The present paper would like to put stress on the new localities for the country, including Sarnena Gora, for future tardigrade research.

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